

In the Specification:

[0162] Additional experiments were done to determine the effects of IGF-I plasmid mediated supplementation using a construct that stimulates the secretion of the transgene product into the general circulation on MyoD expression after a nerve injury. The sciatic nerve of ICR-I female mice was crushed mid thigh. Afterwards, 120 µg of pSP-IGF-I-GH3'UTR (pAV2002 - SEQID No.: 2) , diluted in distilled water was injected in the tibialis anterior in a volume of 30 µl. The muscle was then electroporated at 220V/cm, 20 msec, and 3 pulses, positive and reverse polarity, each. In control mice, 30 µl of distilled water was injected into injured muscle and electroporated as above. After 3 days, the muscle of IGF-I and water-injected mice was analyzed with CELISA. The results shown in Figure 6 indicate that IGF-I plasmid mediated supplementation using a construct that stimulates the secretion of the transgene product into the general circulation does not affect MyoD myogenin expression in the treated muscle.

In the Claims:

1. (Withdrawn) An isolated nucleic acid expression construct comprising:
 - a myogenic promoter;
 - a nucleic acid sequence encoding an insulin-like growth factor I (“IGF-I”) or functional biological equivalent thereof; and
 - a 3' untranslated region (3'UTR);

wherein the isolated nucleic acid expression construct is substantially free from a viral backbone; the myogenic promoter, the nucleic acid sequence encoding IGF-I, and the 3'UTR are operably linked; and the isolated nucleic acid expression construct has an *in vivo* expression activity for the encoded IGF-I or functional biological equivalent thereof in a tissue of a subject.
2. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein the myogenic promoter comprises transcriptional loci from a family of MEF-1, MEF-2, TEF-1, SRE or SP.

3. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein the myogenic promoter comprises a nucleic acid sequence that is at least 85% identical to SEQID No.: 3.
4. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein the encoded IGF-I or functional biological equivalent thereof has an amino acid sequence that is at least 85% identical to SEQID No: 4.
5. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein the encoded IGF-I or functional biological equivalent thereof has an amino acid sequence of SEQID No: 4, or SEQID No.:4 with conservative amino acid substitutions.
6. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein the 3'UTR comprises a nucleic acid sequence that is at least 85% identical to SEQID No.: 5 from a skeletal alpha actin gene, or at least 85% identical to SEQID No.: 6 from a human growth hormone gene.
7. (Withdrawn) The isolated nucleic acid expression construct of claim 1, further comprising a transfection-facilitating vector system.
8. (Withdrawn) The isolated nucleic acid expression construct of claim 7, wherein the transfection-facilitating vector system is a plasmid, a viral vector, a liposome, or a cationic lipid.
9. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein a construct nucleic acid sequence is at least 90% identical to SeqID#1, or a degenerate variant of SEQID#1.
10. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein a construct nucleic acid sequence is at least 90% identical to SeqID#2, or a degenerate variant of SEQID#2.

11. (Withdrawn) The isolated nucleic acid expression construct of claim 1, wherein a construct nucleic acid sequence is Seq. ID No. 1 or Seq. ID No. 2.
12. (Withdrawn) The isolated nucleic acid expression construct of claim 1, further comprising a transfection-facilitating polypeptide.
13. (Withdrawn) The isolated nucleic acid expression construct of claim 12, wherein the transfection-facilitating polypeptide comprises a charged polypeptide.
14. (Withdrawn) The isolated nucleic acid expression construct of claim 11, wherein the transfection-facilitating polypeptide comprises poly-L-glutamate.
15. (Withdrawn) An isolated nucleic acid comprising a sequence at least 95% identical to SeqID#1, or a degenerate variant of SEQID#1.
16. (Withdrawn) An isolated nucleic acid comprising a sequence at least 95% identical to SeqID#2, or a degenerate variant of SEQID#2.
17. (Currently Amended) A method for stimulating angiogenesis, ~~or stimulating myogenesis, or elevating levels of an angiogenic factor, or stimulating endogenous production of an angiopoietin, or treating a muscular or vascular complications of diabetes~~ in a subject, comprising:
injecting into a muscle tissue of the subject an isolated nucleic acid expression construct;
wherein;
the muscle tissue comprises cells; and
the isolated nucleic acid expression construct comprises:
a myogenic promoter;

a nucleic acid sequence encoding an insulin-like growth factor I ("IGF-I") or functional biological equivalent thereof, wherein the IGF-I or functional biological equivalent is capable of binding to an IGF-I receptor ("IGF-IR"); and

a 3' untranslated region (3'UTR);

wherein the isolated nucleic acid expression construct is substantially free from a viral backbone; the myogenic promoter, the nucleic acid sequence encoding IGF-I, and the 3'UTR are operably linked; ~~and the isolated nucleic acid expression construct has an in vivo expression activity for the encoded IGF-I or functional biological equivalent thereof in the tissue of the subject.~~

thereby delivering to the cells of the muscle tissue of the subject the isolated nucleic acid expression construct that enables an in vivo expression activity for the encoded IGF-I or functional biological equivalent thereof stimulating angiogenesis in the tissue of the subject.

18. (Currently Amended) The method of claim 17, wherein further comprising selecting the myogenic promoter comprises a nucleic acid sequence that is at least 85% identical to SEQID No.: 3 and retains a myogenic promoter activity.

19. (Currently Amended) The method of claim 17, wherein further comprising selecting the encoded IGF-I or functional biological equivalent thereof has an amino acid sequence that is at least 85% identical to SEQID No: 4 and retains the function of inducing angiogenesis in the tissue of the subject.

20. (Currently Amended) The method of claim 17, wherein further comprising selecting the encoded IGF-I or functional biological equivalent thereof has an amino acid sequence of SEQID No: 4, or SEQID No.:4 with conservative amino acid substitutions and retains the function of inducing angiogenesis in the tissue of the subject.

21. (Currently Amended) The method of claim 17, wherein further comprising selecting the 3'UTR comprises a nucleic acid sequence that is at least 85% identical to SEQID No.: 5 from a skeletal alpha actin gene, or at least 85% identical to SEQID No.: 6 from a human growth hormone gene and retains 3'UTR activity.
22. (Currently Amended) The method of claim 17, further comprising: mixing the isolated nucleic acid expression construct with a transfection-facilitating ~~vector~~ system before delivering the isolated nucleic acid expression construct into the tissue of the subject.
23. (Currently Amended) The method of claim 22, wherein the transfection-facilitating ~~vector~~ system is, ~~a viral vector~~, a liposome, or a cationic lipid.
24. (Currently Amended) The method of claim 17, wherein further comprising selecting a construct nucleic acid sequence that is at least 90% identical to SeqID#1, or a degenerate variant of SEQID#1 and retains the function of inducing angiogenesis in the tissue of the subject.
25. (Withdrawn) The method of claim 17, wherein a construct nucleic acid sequence is at least 90% identical to SeqID#2, or a degenerate variant of SEQID#2.
26. (Currently Amended) The method of claim 17, wherein a construct nucleic acid sequence is Seq. ID No. 1 ~~or Seq. ID No. 2.~~
27. (Original) The method of claim 17, further comprising mixing the isolated nucleic acid expression construct with an effective concentration of a transfection-facilitating polypeptide before delivering the isolated nucleic acid expression construct into the tissue of the subject.
28. (Original) The method of claim 27, wherein the transfection-facilitating polypeptide comprises a charged polypeptide.

29. (Original) The method of claim 27, wherein the transfection-facilitating polypeptide comprises poly-L-glutamate.
30. (Original) The method of claim 17, further comprising electroporating the tissue after the nucleic acid expression construct has been delivered into the tissue of the subject.
31. (Original) The method of claim 17, wherein the nucleic acid expression construct is delivered into the tissue of the subject via a single administration.
32. (Currently Amended) The method of claim 17, wherein the cells of the subject are somatic cells, stem cells, ~~or germ cells~~.
33. (Original) The method of claim 17, wherein the cells of the tissue are diploid cells.
34. (Original) The method of claim 17, wherein delivering the nucleic acid expression construct initiates expression of the encoded IGF-I or functional biological equivalent thereof.
35. (Original) The method of claim 34, wherein the encoded IGF-I or functional biological equivalent thereof is expressed in tissue specific cells of the subject.
36. (Original) The method of claim 35, wherein the tissue specific cells of the subject comprise muscle cells.
37. (Original) The method of claim 34, wherein the encoded IGF-I is a biologically active polypeptide; and the encoded functional biological equivalent of IGF-I is a polypeptide that has been engineered to contain a distinct amino acid sequence while simultaneously having similar or improved biological activity when compared to the IGF-I polypeptide.
38. (Original) The method of claim 17, wherein the subject is a human, a pet animal, a farm animal, a food animal, or a work animal.

39. (Withdrawn) The method of claim 17, wherein the angiogenic factor comprises a vascular endothelial growth factor (“VEGF”) having an amino acid sequence that is at least 85% identical to SEQID No: 7.
40. (Withdrawn) The method of claim 17, wherein the angiogenic factor comprises a vascular endothelial growth receptor (“VEGF receptor”).